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Crystal Structure of *Agaricus bisporus* Mushroom Tyrosinase

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Supplementary Information belonging to

Wangsa T. Ismaya, Henriëtte J. Rozeboom, Amrah Weijn, Jurriaan J. Mes, Fabrizia Fusetti, Harry J. Wichers and Bauke W. Dijkstra. Crystal structure of *Agaricus bisporus* mushroom tyrosinase - Identity of the tetramer subunits and interaction with tropolone.

Fig. S1. *A. bisporus* tyrosinase tetramer in the asymmetric unit of a) space group $P2_12_12$ (dimensions $\sim 139 \times 97 \times 59$ Å), and b) space group $P2_1$ (dimensions $\sim 107 \times 87 \times 84$ Å). The H subunits are coloured red and green, and the L subunits are in light blue and purple. The black balls represent the positions of the bound holmium ions (see main text for explanation).

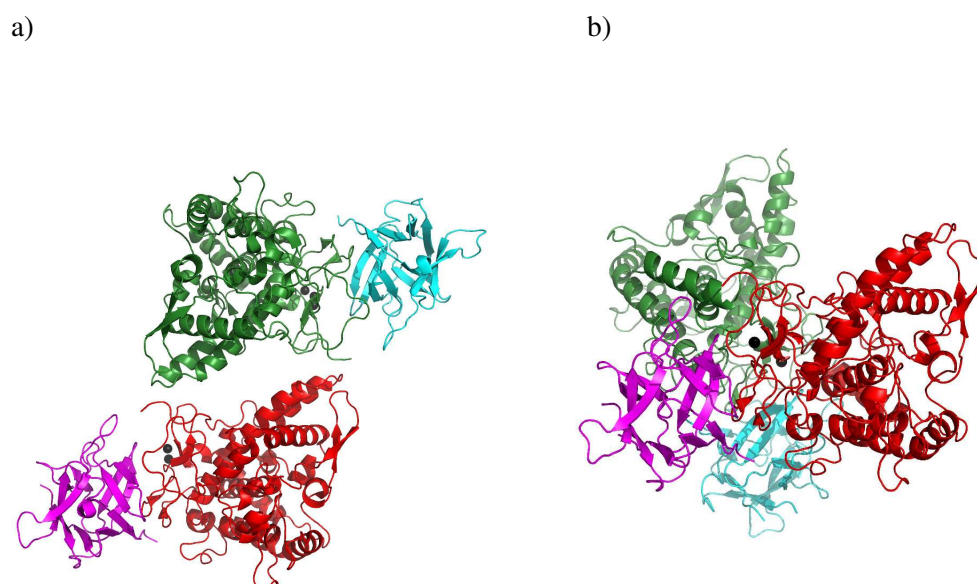


Fig. S2. Structure-based sequence alignment of tyrosinase-related type 3 copper proteins. The aligned structures and PDB database accession numbers are *AbTyr*, *A. bisporus* tyrosinase (PPO3, this article; PDB 2y9w); *BmTyr*, *B. megaterium* tyrosinase (PDB 3nm8 (1)); *ScTyr*, *S. castaneoglobisporus* tyrosinase (PDB 1wx2 (2)); *IbCox*, *I. batata* catechol oxidase (PDB 1bt1 (3)) and *OdHcy*, *Octopus dofleini* hemocyanin (PDB 1js8 (4)). Structural alignment was performed using the DALI server. The secondary structure elements are shown as squiggles (α -helix and 3_{10} -helix), black arrows (β -strands) and strict β -turns as **TT** letters at the top of

[illegible]

Fig. S3. Stereo figure of a superposition of *A. bisporus* tyrosinase (green; PDB 2y9w (this article)), *B. megaterium* tyrosinase (orange; PDB 3nq1 (1)), *S. castaneoglobisporus* tyrosinase (magenta; PDB 1wx2 (2)). Copper ions are in light brown and the inhibitor kojic acid from the *B. megaterium* tyrosinase structure is shown in orange.

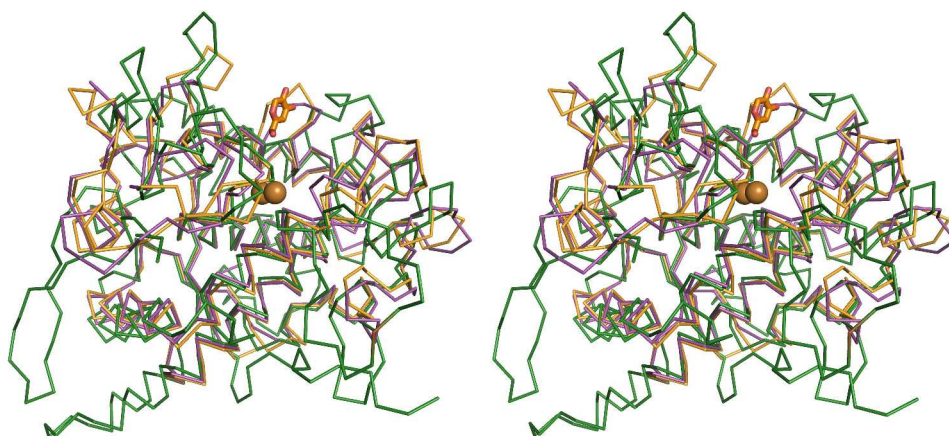
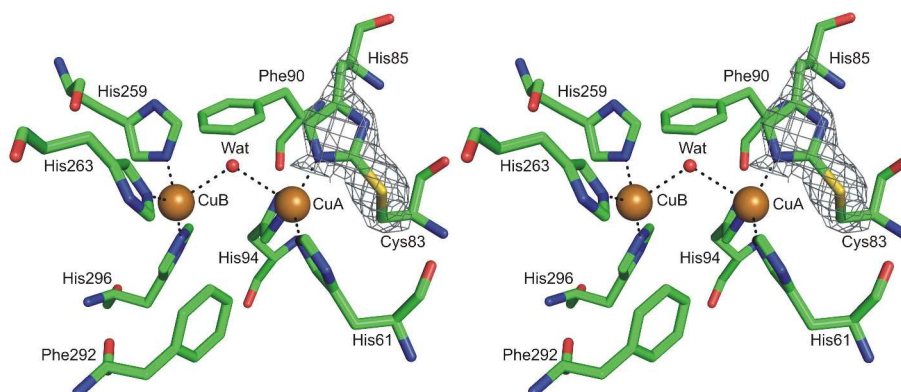


Fig. S4. Stereo figure of the geometry of the binuclear copper-binding site in space group $P2_12_12_1$. The ligands are colored according to atom type (green, blue, red, yellow for carbon, nitrogen, oxygen, sulfur, respectively). The brown spheres are the Cu-A and Cu-B copper ions, and the red sphere is a bound water molecule/hydroxyl ion. The electron density for the covalent thioether bond between the Cys83 S γ and His85 C ϵ 1 atoms is contoured at 2.0 σ and is shown in black mesh.



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